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Nailfold capillaroscopy: A sensitive method for evaluating microvascular involvement in children with SARS-CoV-2 infection

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ABSTRACT

Objectives: The hyperinflammatory state and the viral invasion may result in endothelial dysfunction in SARS-CoV-2 infection. Although a method foreseeing microvascular dysfunction has not been defined yet, studies conducted in patients diagnosed with COVID-19 have demonstrated the presence of endotheliitis. With this study, we aimed to investigate the microvascular circulation in patients diagnosed with COVID-19 and multi-system inflammatory syndrome in children (MIS-C) by nailfold videocapillaroscopy (NVC).

Methods: Thirty-one patients with SARS-CoV-2 infection, 25 of whom were diagnosed with COVID-19 and 6 with MIS-C and 58 healthy peers were included in the study. NVC was performed in eight fingers with 2 images per finger and 16 images were examined for the morphology of capillaries, presence of pericapillary edema, microhemorrhage, avascular area, and neoangiogenesis. Capillary length, capillary width, apical loop, arterial and venous width, and intercapillary distance were measured from three consecutive capillaries from the ring finger of the non-dominant hand.

Results: COVID-19 patients showed significantly more capillary ramification ($p < 0.001$), capillary meandering ($p = 0.04$), microhemorrhage ($p < 0.001$), neoangiogenesis ($p < 0.001$), capillary tortuosity ($p = 0.003$). Capillary density ($p = 0.002$) and capillary length ($p = 0.002$) were significantly lower in the patient group while intercapillary distance ($p = 0.01$) was significantly longer compared with healthy volunteers. Morphologically, patients with MIS-C had a higher frequency of capillary ramification and neoangiogenesis compared with COVID-19 patients ($p = 0.04$).

Conclusion: Abnormal capillary alterations seen in COVID-19 and MIS-C patients indicate both similar and different aspects of these two spectra of SARS-CoV-2 infection and NVC appears to be a simple and non-invasive method for evaluation of microvascular involvement.

1. Introduction

The coronavirus (SARS-CoV-2) pandemic, known as COVID-19 has spread all over the world in a short period of time and caused the death of more than 2 million people to date (World Health Organization (WHO).org [Internet], n.d.). Although in severe cases, it mainly progresses as a serious lung disease such as pneumonia or acute respiratory distress syndrome (ARDS), numerous extrapulmonary manifestations due to systemic hyperinflammation associated with COVID-19 have

been described (Salton et al., 2020; Meduri et al., 2020). The literature enlightening organ-specific pathophysiologies have started to emerge expeditiously (Gupta et al., 2020).

It is now clear that the entry of the virus into the cell is dependent on angiotensin-converting enzyme 2 (ACE2) and transmembrane serine protease 2 receptors (Hoffmann et al., 2020). Postmortem examinations revealed endothelial inflammation and endothelial cell (EC) death, accordingly endotheliitis might play a crucial role in microvascular damage in COVID-19 (Varga et al., 2020). The levels of von Willebrand

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Factor (VWF) and p-selectin were significantly high in some patient series, indicating microvascular inflammation and thrombosis. The resulting vascular involvement also increases the release of cytokines such as interleukin 6, interleukin (IL)-1 β , and tumor necrosis factor- α (TNF- α), thus initiating the cycle of vascular damage and tissue inflammation that contribute to the prolonged hyperinflammatory state and hypercoagulability (Lowenstein and Solomon, 2020; Colantuoni et al., 2020). All these results call attention to endothelial leakage and dysfunction as the originator of ARDS, and systemic inflammation observed in COVID-19 (Teuwen et al., 2020).

The SARS-CoV-2 viral load is greater in children than adults and self-limited, chilblain-like acral purpuric lesions are the best evidence of that aspect (Colantuoni et al., 2020). It was depicted that children experience viral tropism in their ECs during acute infection with viremia, even if the disease course remains subclinical (Heald-Sargent et al., 2020; Hernandez and Bruckner, 2020). The demonstration of SARS-CoV-2 in ECs of skin biopsies by immunohistochemistry and electron microscopy confirm these lesions as part of the spectrum of COVID-19 (Colmenero et al., 2020).

Since it is not clear whether the immunological mechanism of COVID-19 inducing hyperinflammation in children is akin to the mechanisms in adults, the disease course may be distinctive during childhood. In April 2020, COVID-19 reports of children with clinical and laboratory features similar to Kawasaki disease and toxic shock syndrome begin to come out (Verdoni et al., 2020; Cook et al., 2020). Afterwards, a unique syndrome with fever and hyperinflammation affecting not only the skin and heart but also the gastrointestinal, neurological, and respiratory systems were defined and then currently named as multisystem inflammatory syndrome in children (MIS-C) (Henderson and Yeung, 2021). While evaluating patients attentively, alterations of the microcirculation and endothelial structures were demonstrated in adult studies, however evidence of microvascular dysfunction in children with COVID-19 or MIS-C is scarce (Rovas et al., 2021; Favaron et al., 2021; Goshua et al., 2020).

Capillaroscopic examination of the nailfold is a feasible method for monitoring the microvascular circulation, and the capillaries of the nail bed lying parallel to the skin surface may be visualized non-invasively (Smith et al., 2020; Ruaro et al., 2018). Nailfold capillaroscopy (NFC) is a routine method used in rheumatology practice to investigate connective tissue diseases and to distinguish between primary and secondary Raynaud's phenomenon, and moreover, abnormal nailfold capillaroscopy (NFC) is included in the 2013 American College of Rheumatology (ACR)/European League Against Rheumatism (EULAR) classification criteria for systemic sclerosis (van den Hoogen et al., 2013). However, markers of endothelial damage may be elevated in patients with Raynaud's phenomenon who subsequently develop systemic scleroderma or other connective tissue diseases, even in the absence of capillaroscopic abnormalities (Gualtierotti et al., 2017).

The objective of our study is to evaluate nailfold capillaroscopic examination of children diagnosed with COVID-19 and MIS-C, and to compare the images with healthy children's capillaroscopic findings, for investigating the microvascular circulation alterations in children with COVID-19.

2. Materials AND methods

2.1. Patients data and study design

This study included children diagnosed and followed-up with SARS-CoV-2 infection at Istanbul Faculty of Medical School by pediatric emergency, pediatric infectious diseases, pediatric rheumatology, and pediatric intensive care units.

Sociodemographic data and disease-related information (symptoms at COVID-19 onset, disease duration, clinical features, radiologic findings, laboratory evaluations, applied treatment modalities, comorbidities, and complications attributed to COVID-19) were assessed from the

files of the patients. Any individual aged between 5 and 18 years with SARS-CoV-2 infection confirmed by reverse transcription-polymerase chain reaction (RT-PCR), suggestive chest imaging, or positive SARS-CoV-2 antibodies in the last three months was eligible for inclusion. MIS-C was defined as; an individual aged <21 years presenting with fever, laboratory evidence of inflammation, and evidence of clinically severe illness requiring hospitalization, with multisystem (≥ 2) organ involvement (cardiac, renal, respiratory, hematologic, gastrointestinal, dermatologic, or neurological) with no alternative plausible diagnosis and be positive for current or recent SARS-CoV-2 infection by RT-PCR, serology or antigen test; or COVID-19 exposure within 4 weeks before the onset of symptoms (Centers for Disease Control and Prevention, n. d.).

2.2. Nailfold videocapillaroscopy (NVC) examination

NVC was performed both in the patient group and control group. The control group consists of age and sex-matched healthy peers without a prior history of chronic disease or smoking and traumatization of hands. Children with a diagnosis of chronic rheumatic disease and Raynaud's syndrome were not included in the patient group. Imaging sessions were carried out after 20 min acclimatization at a room temperature of 23 °C (Dolezalova et al., 2003). The NVC was carried out on eight fingers (excluding thumbs) on both hands of each patient, after a drop of immersion oil placed on the nailfold bed to improve resolution. NVC images were captured using the Dino-Lite CapillaryScope 200 Pro/MEDL4N Pro capillaroscopy device at 200 \times magnification. All fingers except the thumbs were examined paying greater attention to the ring finger of the non-dominant hand for the presence of any abnormality bilaterally and two images from eight fingers were obtained from both the study and control groups.

The images were assessed at the end of the examination by the same executing physician in a blinded way without having any knowledge about the clinical features of the patients examined. The morphology (*i. e.* capillary tortuosity, capillary crossing, enlarged capillary, giant capillary, capillary meandering, and branched capillary) of the capillary, presence of pericapillary edema, microhemorrhage, avascular areas, and neoangiogenesis (bushy capillaries, pathologic branching, capillary ramifications) were the parameters evaluated during the capillaroscopic examination of eight fingers and pathologic findings were reported from 16 images. These parameters were assessed as present or absent, and the presence of signs in at least two fingers was recorded as capillary abnormality in both groups. An increase in capillary diameter (arterial, venous or apical loop) from 20 μ m to 50 μ m was assessed as enlarged capillary, and a homogeneously enlarged loop with a diameter ≥ 50 μ m was assessed as giant capillary (Sulli et al., 2008). An intercapillary distance greater than 500 μ m in the distal capillary row was evaluated as an avascular area (Cutolo et al., 2000). The meandering capillary was defined as the limbs crossed upon themselves or with other several times and bushy capillary was the small multiple buds originated from the limbs (Andrade et al., 1990).

The capillary tortuosity and capillary crossing were defined as follows: the absence of alterations (0), less than 50% of examined capillaries altered (1), more than 50% of examined capillaries altered (2). When more than 50% of the capillaries were tortuous, this was considered to be a major abnormality (Ingegnoli et al., 2005). The reduced capillary density was evaluated by comparison with the healthy controls. The measurements were made at three consecutive capillaries from the ring finger of the non-dominant hand for capillary length, capillary width, apical loop, arterial and venous width, and distance between capillaries. Finally, for each child in the patient and control group, it was checked whether there was a scleroderma pattern in the NVC evaluations (Cutolo et al., 2000; Maricq, 1981).

All participants gave informed written consent for participation in the study and the study was conducted according to Good Clinical Practice guidelines and the Declaration of Helsinki, with necessary

approvals from the local Ethical Committee (approval number: E-29624016-050.99-107885).

The SPSS version 21.0 (SPSS, Inc., Chicago, Illinois) is used for statistical analysis. The variables were investigated using visual (histogram, probability plots) and analytic methods (Kolmogorov Smirnov/Shapiro-Wilk's test) to determine whether or not they were normally distributed. Descriptive analyses were presented using proportions, mean, standard deviation (SD), medians, minimum, and maximum values as appropriate.

The Student *t*-test and Mann-Whitney *U* test were used to compare non-normally distributed variables between the two groups. A value of $P < 0.05$ was considered statistically significant.

3. Results

3.1. Baseline characteristics of patients and control group

A total of 31 patients were enrolled in the study. Among them, 14 (45.1%) were female and 17 (54.9%) were male. The median age of patients was 12 (5–18) years. Fifty-eight age and sex-matched healthy volunteers were included in the study as a control group. Of 58 healthy individuals, 28 (48.2%) were female and 30 (51.8%) were male. The median age of the control group was 12 (5–18) years.

In the patient cohort, 25 (80.6%) were diagnosed with COVID-19, and 6 (19.4%) were with MIS-C. Fourteen (45.2%) patients had a concomitant disease (asthma = 2, atopic dermatitis = 1, familial Mediterranean fever = 3, hypothyroidism = 1, congenital adrenal hyperplasia = 1, chronic immune thrombocytopenic purpura = 1, history of liver transplant = 2, cyclic neutropenia = 1, cerebrospinal fluid leak = 1, schizencephaly = 1) and 19 (61.3%) patients in the study cohort had a history of COVID-19 contact. Among patients diagnosed with COVID-19, 19 (76%) had presented with fever and 14 (56%) had sore throat. The other accompanying features were as follows, vomiting and nausea in seven (28%), neurologic involvement in seven (28%), myalgia in seven (28%), arthralgia in three (12%) headaches in two (8%), cough in two (8%), cardiac involvement in two (8%) patients and diarrhea in one (4%) patient. There were no skin and/or vasculitic lesions in our patient group. None of them required oxygen support, mechanical ventilation, inotropic support, or hospitalization in the intensive care unit. SARS-CoV-2 RT-PCR positivity was present in 23 (92%) patients and the remaining two (8%) patients had positive findings compatible with COVID-19 on thorax CT. Among these 25 patients, three (12%) were treated with antiviral agents, and two (8%) received enoxaparin. The median duration of hospitalization was 5.5 (4–12) days. Nineteen patients were followed in the emergency department for less than 24 h. All patients recovered without any complications.

Six (19.4%) patients of the cohort were diagnosed with MIS-C. All attended to the hospital with fever and cardiovascular signs and symptoms (tachycardia, hypotension, shock), five (83.3%) patients had gastrointestinal symptoms (nausea-vomiting, abdominal pain), four (16.7%) had hematologic involvement (lymphopenia, thrombocytopenia), and two (33.3%) had cough. Furthermore, all patients required both supportive care with oxygen and inotropic agents, and two (33.3%) were treated with non-invasive ventilation. All patients received anticoagulant drugs (enoxaparin = 5 and acetylsalicylic acid = 1), five (83.3%) patients were treated with pulse methylprednisolone (30 mg/kg/day), and two with intravenous immunoglobulin (IVIG) (2 g/kg). Patients with MIS-C were all positive for the SARS-CoV-2 antibody. The median duration of hospitalization was 5 (4–7) days and they recovered without any complications.

The laboratory parameters of patients were depicted in [Table I](#).

3.2. Nailfold videocapillaroscopy examination of patient and control group

NVC examination was performed to all of the patients within a

Table I

The laboratory parameters of patients with COVID-19 and Multisystem Inflammatory Syndrome in Children (MIS-C).

Parameters	Patients with COVID-19 (n = 25)	Patients with MIS-C (n = 6)
Complete blood cell counts		
White blood counts (cells/ μ L) ^a	5400 (2600–11,300)	9950 (3800–18,700)
Hemoglobin, (g/dL) ^a	13.1 (9.9–15.4)	11.1 (10–13.9)
Lymphocyte counts (cells/ μ L) ^a	1250 (500–3500)	900 (500–2400)
Lymphopenia, n (%)	15 (60%)	4 (66.7%)
Platelet counts (cells/ μ L) ^a	261,500 (200,000–344,000)	173,000 (102,000–297,000)
Thrombocytopenia, n (%)	0 (0)	3 (50)
Inflammatory markers		
C-reactive protein (CRP) (mg/dL) ^a	5.5 (0.24–59)	128 (33.7–289)
Elevated CRP, n (%)	11 (44)	6 (100)
Ferritin (ng/mL) ^a	56 (5–193)	474 (65–1137)
Elevated ferritin, n (%)	0 (0)	4 (66.7)
Procalcitonin (ng/mL) ^a	0.07 (0.02–0.21)	5.1 (0.86–11.5)
Elevated procalcitonin, n (%)	0 (0)	6 (100)
Coagulation tests		
D-dimer (ng/L) ^a	330 (230–2020)	2610 (1460–148060)
Elevated D-dimer, n (%)	4 (16)	6 (100)
International normalized ratio	1 (0.69–1.36)	1.23 (1–1.5)
Activated prothrombin time (sec)	29 (19.8–34.8)	27.4 (10–33)
Prothrombin time (sec)	13.8 (7.8–17.7)	46 (13.2–20)
Fibrinogen (mg/dL)	323 (255–508)	512 (316–796)
Cardiac markers		
NT-pro-BNP (pg/mL) ^a	32 (10–547)	3913 (811–33,250)
Elevated NT-pro-BNP, n (%)	1 (4)	6 (100)
Other biochemical markers		
Urea (mg/dL) ^a	21 (14.4–36)	24 (15.8–44.5)
Creatinine (mg/dL) ^a	0.68 (0.46–0.9)	0.43 (0.3–1.3)
Albumin (g/dL) ^a	4.8 (4.4–5.5)	3.1 (2.5–4.4)
Aspartate aminotransferase (U/L) ^a	21 (13.9–34.6)	33 (12.6–49)
Alanine aminotransferase (U/L) ^a	11.9 (7.6–30)	40 (11.3–62)

COVID-19, coronavirus disease 2019; MIS-C, multisystem inflammatory syndrome in children.

^a Data expressed as median (minimum–maximum).

median 73 (8–104) days after diagnosis. A total of 62 images were obtained from 31 patients and 116 images from 58 healthy children. Two hundred sixty-seven capillary measurements were made. Patients had significantly lower capillary density compared to healthy volunteers ($p = 0.002$). The capillary length was significantly lower in the patient group while the intercapillary distance was significantly longer in the patient group as compared to healthy volunteers. When we assessed the capillary morphology, the patients group had a higher frequency of capillary branching and meandering, bushy capillaries, microhemorrhages, and neoangiogenesis. Neither the patient nor the control group had avascular areas and giant capillaries detected in the NVC examinations. ([Table II](#)). The comparison of NVC findings of patients with COVID-19 and patients with MIS-C was summarized in [Table III](#). Morphologically, patients with MIS-C had a higher frequency of capillary ramification and neoangiogenesis. The representative pictures of capillary alterations of children diagnosed with SARS-CoV-2 infection and capillaroscopic changes seen in the healthy controls were shown in [Fig. 1](#).

The patients with abnormal findings (capillary branching, capillary meandering, bushy capillary, capillary ramification, microhemorrhage,

Table II
Nailfold videocapillaroscopy (NVC) examination of patient and control group.

Parameters	Patients group (n = 31)	Control group (n = 58)	P value
Capillary density ^a	6 (5–10)	7 (4–10)	0.002
Capillary length ^a (µm)	300 (104–555)	327 (83–750)	0.002
Arterial width ^a (µm)	10 (6–25)	11 (6–19)	0.91
Venous width ^a (µm)	13 (6–31)	13.5 (7–26)	0.09
Apical loop width ^a (µm)	15 (7–39)	16 (8–31)	0.58
Intercapillary distance ^a (µm)	129 (37–341)	106 (39–319)	0.01
Capillary tortuosity, n (%)			0.003
<50%	4 (12.9)	29 (50)	
>50%	5 (16.1)	0	
None	22 (71)	29 (50)	
Crossing capillary, n (%)			0.34
<50%	15 (48.4)	26 (44.8)	
>50%	1 (3.2)	0	
None	15 (48.4)	32 (55.2)	
Dilated (enlarged) capillary, n (%)	12 (38.7)	18 (31.1)	0.46
Giant capillary, n (%)	0 (0)	0 (0)	NA
Avascular area, n (%)	0 (0)	0 (0)	NA
Capillary branching, n (%)	8 (25.8)	0 (0)	<0.001
Capillary meandering, n (%)	3 (9.6)	0 (0)	0.04
Bushy capillary, n (%)	8 (25.8)	0 (0)	<0.001
Capillary ramification, n (%)	9 (29)	0 (0)	<0.001
Microhemorrhage, n (%)	7 (22.5)	0 (0)	<0.001
Pericapillary edema, n (%)	2 (6.4)	0 (0)	0.11
Neoangiogenesis, n (%)	9 (29)	0 (0)	<0.001

^a Data expressed as median (minimum–maximum).

Table III
Comparison of nailfold capillaroscopic findings between patients with COVID-19 and patients with Multisystem Inflammatory Syndrome in Children (MIS-C).

Parameters	Patients with COVID-19 (n = 25)	Patients with MIS-C (n = 6)	P value
Capillary density ^a	6 (5–10)	6 (5–8)	0.67
Capillary length ^a (µm)	294 (104–555)	319 (107–486)	0.82
Arterial width ^a (µm)	10 (6–25)	10 (6–20)	0.29
Venous width ^a (µm)	13 (9–31)	11 (6–22)	0.14
Apical loop width ^a (µm)	16 (10–39)	14 (7–24)	0.05
Capillary distance ^a (µm)	37 (24–84)	31 (21–50)	0.09
Capillary tortuosity, n (%)			0.95
<50%	3 (12)	1 (16.6)	
>50%	4 (16)	1 (16.6)	
None	18 (72)	4 (66.8)	
Crossing capillary, n (%)			0.04
<50%	11 (44)	4 (66.8)	
>50%	0	1 (16.6)	
None	14 (56)	1 (16.6)	
Dilated capillary, n (%)	12 (48)	0 (0)	0.03
Giant capillary, n (%)	0 (0)	0 (0)	NA
Avascular area, n (%)	0 (0)	0 (0)	NA
Capillary branching, n (%)	6 (24)	2 (33.3)	0.63
Capillary meandering, n (%)	2 (8)	1 (16.6)	0.48
Bushy capillary, n (%)	5 (20)	3 (50)	0.16
Capillary ramification, n (%)	4 (16)	4 (66.8)	0.04
Microhemorrhage, n (%)	6 (24)	1 (16.6)	0.58
Pericapillary edema, n (%)	1 (4)	1 (16.6)	0.35
Neoangiogenesis, n (%)	5 (20)	4 (66.8)	0.04

COVID-19, coronavirus disease 2019; MIS-C, multisystem inflammatory syndrome in children.

^a Data expressed as median (minimum–maximum).

precapillary edema, neoangiogenesis, and microhemorrhage) had higher levels of C-reactive protein (CRP) and D-dimer (CRP; 16.4 vs 2.2, $p = 0.04$ and D-dimer; 900 vs 340, $p = 0.04$).

4. Discussion

SARS-CoV-2 infection by affecting the vascular system and coagulation properties of the blood precipitates the injury along the vascular walls and clot formation in both large and microscopic blood vessels. The research regarding microvascular and endothelial injury may engender a fundamental explanation for the pathophysiological mechanisms of COVID-19. The endothelial injury, indeed, appears to be the key pathophysiologic factor leading to multi-organ failure and even to death (Li et al., 2020). ACE2 mediates SARS-CoV-2 entry into the host cell, and it is a predominant receptor on the EC membrane. ECs play a critical role in the inflammatory and disseminated coagulation processes reported, which culminate in a higher risk of thromboembolic, cardiovascular, and cerebrovascular complications in these patients (Barbosa et al., 2021). Due to the recent evidence for the presence of microvascular damage in patients with COVID-19, evaluating the nail bed capillaries of children diagnosed with COVID-19 may shed light on the EC injury.

To the best of our knowledge, this observational study is the first one defining NVC findings in children diagnosed with COVID-19 and MIS-C. When we compared the NVC findings of healthy children with patients; we noticed that the frequency of abnormal findings such as microhemorrhage, precapillary edema, capillary meandering, capillary branching, bushy capillary, capillary ramification, and neoangiogenesis were significantly more common in the patient group. Furthermore, patients had significantly lower capillary density, capillary length, and higher intercapillary distance as compared to healthy volunteers. Patients with higher CRP and D-dimer levels were more likely to have pathological NVC findings.

During NVC examination, 25 patients were at the recovery phase of COVID-19 with a median follow-up duration of 83 (20–104) days post infection and 6 patients with MIS-C were examined at a median period of 16 (8–49) days following discharge from the hospital. When patients diagnosed with COVID-19 and MIS-C were compared through NVC findings; capillary ramification and neoangiogenesis were significantly more common in the MIS-C group, however, enlarged capillaries were more common in the COVID-19 patients. According to the clues from the current literature, the pathogenesis of severe COVID-19 appears to be associated with the systemic inflammation and thrombosis observed in infected patients (Klok et al., 2020). Patients with severe COVID-19 may have laboratory findings consistent with a cytokine storm, widespread thrombosis, and fibrinolysis. They are characterized with very high levels of erythrocyte sedimentation rate (ESR), CRP, ferritin, interleukins (IL-1 β , IL-1RA, IL-2), and D-dimer (Zhou et al., 2020; Henry et al., 2020; Kermali et al., 2020). Hypoxia, hyperinflammation, and hypercoagulability during the disease course may stimulate neoangiogenesis in the nail bed capillaries and as in our cohort, this may demonstrate why neoangiogenesis is significantly more pronounced in patients with MIS-C. Similarly, Emanule et al., who examined sublingual microcirculation by a hand-held vital microscope based on incident dark field microscopy imaging in COVID-19 patients in the ICU, reported that as a response to hypoxemia, hyperinflammation, and hypercoagulation the number of vessels and functional capillary density in COVID-19 patients were increased in comparison to healthy volunteers (Favaron et al., 2021). In our cohort, even though the capillary density was lower with respect to controls, neoangiogenesis was significantly noticeable in patients diagnosed with COVID-19, chiefly MIS-C. In an autopsy series, Maximillian et al. reported that the amount of new vessel growth (predominantly through a mechanism of intussuscepted angiogenesis) in the lungs of the patients who died from COVID-19 was 2.7 times as high as that in the lungs from patients with influenza (Ackermann et al., 2020).

Coronaviruses show tropism to many cell types in the body including

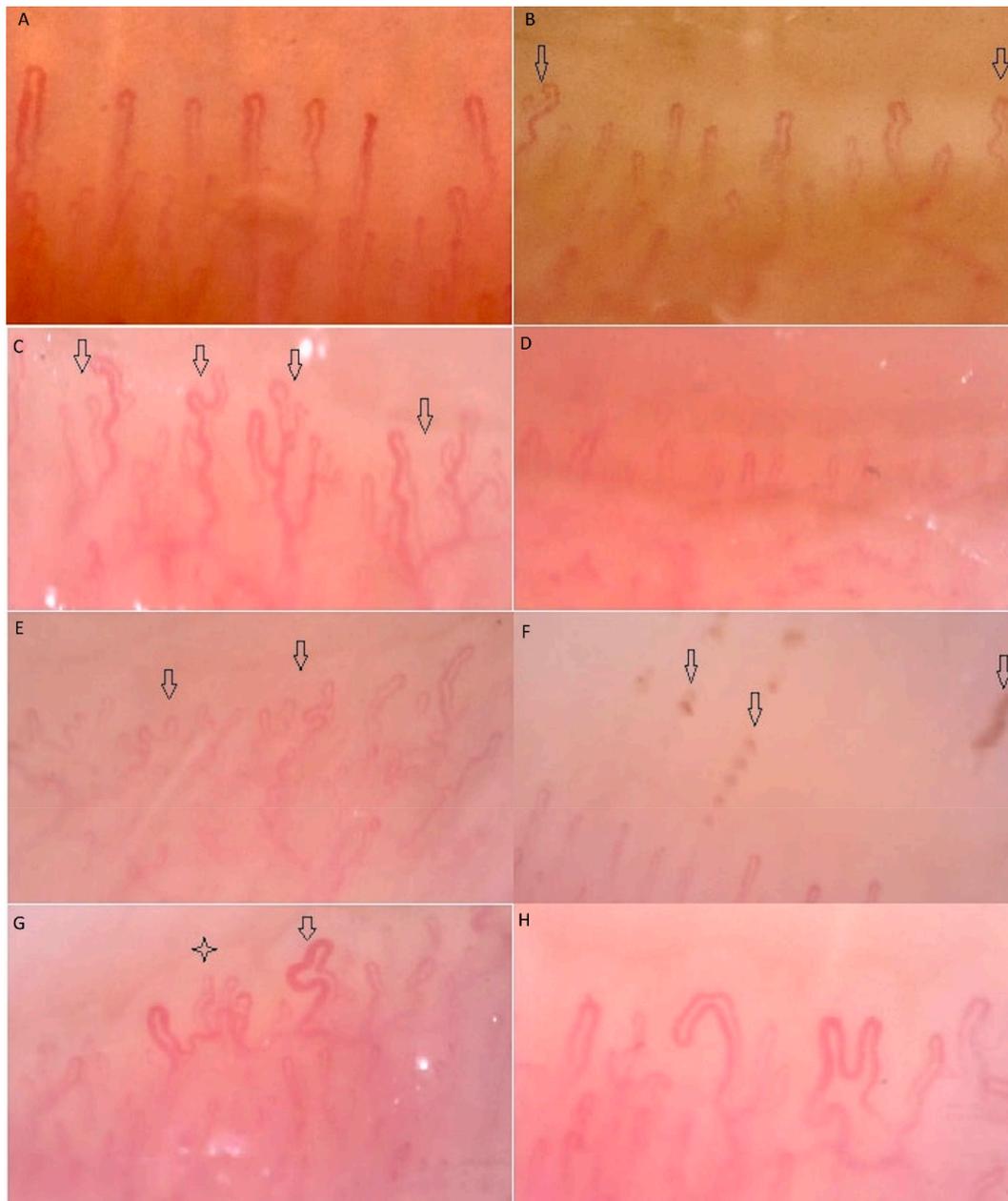


Fig. 1. Normal capillaries in healthy controls and nailfold capillaroscopy abnormalities found in patients with COVID-19 and MIS-C (200× magnification). A) Normal capillaries with a “hairpin” shape in healthy controls; B) normal tortuous capillaries in healthy controls: less than 50% of capillaries, without crossing (arrows); C) capillary ramifications: branching, bushy or coiled capillaries originated from a single capillary (arrows); D) pericapillary edema: capillaries that appear indistinct due to pericapillary edema; E) Neovascularization (arrows); F) capillary micro-hemorrhage: dark mass due to haemosiderin deposit (arrows); G) bushy capillary (four-pointed star) and enlarged capillary with an increase in capillary diameter (20–50 μm) (arrows); H) Microvascular derangement: irregular capillary distribution and orientation with shape heterogeneity of the loops.

epithelial cells lining the respiratory and gastrointestinal tract, ECs, and myocardial cells (V’Kovski et al., 2021). In this cohort, six of the 8 patients with cardiac involvement were diagnosed with MIS-C and two of these patients had coronary involvement, three had valvular insufficiency and three had abnormal echocardiography (coronary artery ectasia, valve regurgitation) findings. All 6 patients had both cardiac involvement and abnormal nail bed capillaroscopy findings. Therefore, we may speculate that COVID-19 appears to progress with vasculitis affecting both small and medium sized vessels, based on the fact that cardiac involvement and small vessel involvement are interrelated in patients with MIS-C diagnosis.

Ingegnoli et al. conducted a study among 118 children diagnosed with a chronic rheumatic disease (juvenile idiopathic arthritis, mixed

connective tissue disease, Raynaud’s phenomenon, systemic lupus erythematosus, systemic sclerosis, and juvenile dermatomyositis), and investigated minor and major pathological changes by performing NVC. In their study, JIA subtypes were examined; although scleroderma pattern was not observed in any of the patients diagnosed with JIA, major abnormalities were mostly observed in the systemic JIA (sJIA) subgroup compared with other subtypes (Ingegnoli et al., 2005). Similar to this study, capillaroscopic abnormalities were more common in patients diagnosed with MIS-C accompanied by systemic hyperinflammation, as in the sJIA subgroup in which inflammatory response of JIA is more prominent.

Although COVID-19 in children has a milder course, MIS-C shows a close similarity with the hyperinflammatory syndromes such as

Kawasaki Disease shock syndrome, toxic shock syndrome, and macrophage activation syndrome (Han and Lee, 2020). Huang et al. assessed cutaneous microcirculation of patients diagnosed with KD by dynamic capillaroscopy and correlated it with coronary artery diameter in these patients. They showed that the capillary morphology was abnormal when compared to controls, with a larger diameter of the arterial and venous limbs, a higher intercapillary distance, and a decrease in the loop numbers of the capillaries. The relationship between peripheral cutaneous vascular change and coronary arterial status observed in this study suggests that dynamic capillaroscopy is a non-invasive method of identifying small vessel changes and, indirectly, identifying coronary arterial abnormalities in KD patients (Huang et al., 2012). Likely, in addition to the evidence of both cardiac and capillary involvement in patients with MIS-C, we demonstrated that COVID-19 patients have a lower capillary density and higher intercapillary distance compared to healthy controls.

Even though there is very limited data regarding capillaroscopic changes in COVID-19, 13 month-old boy diagnosed with MIS-C was reported to have capillary abnormalities (capillary leakage, bizarre capillaries, and irregular ectasia) illustrated by NFC examination (Terrieri et al., 1999). In the first study reporting the capillary abnormalities in 82 adults of whom 28 were in the acute phase and 54 were in the recovery phase of COVID-19 pneumonia, microvascular abnormalities were depicted in both groups. The presence of hemosiderin deposits and precapillary edema were marked in the patients evaluated in the acute phase, while patients who recovered showed prominence of enlarged capillaries, meandering capillaries, and lower capillary density.

In our cohort a disarranged pattern except for minor abnormalities such as capillary tortuosity, capillary crossing, and enlarged capillary were not detected in healthy controls and while a variable range of capillary abnormalities were present in patients diagnosed with COVID-19 and MIS-C, however scleroderma pattern defined according to the classification proposed by Cutolo et al. was not found in any of the patient population (Sulli et al., 2008; Cutolo et al., 2000).

The small population of the cohort is the major limitation nevertheless, this is the only study showing the impact of SARS-CoV-2 infection on microvascular circulation in a pediatric population by NVC. Patients of the COVID-19 and MIS-C group were not distributed evenly for a definitive comparison however, we still assume that the findings of the study are giving physicians an idea about the extent of inflammation and microvascular involvement by SARS-CoV-2 infection. A larger cohort may be more demonstrative for making such a conclusion. As portrayed in the previous publications, symptomatic patients with SARS-CoV-2 infection generally had a concomitant disease, in our cohort nearly half of the affected patients had a concomitant disease as well, while participants from the control group had no co-morbid condition as engendered during the the natural course of the disease (Marchetti, 2020).

5. Conclusion

Children diagnosed with COVID-19 and MIS-C present with several microvascular abnormalities on NVC examination. MIS-C is an emergency phenomenon in which evidence suggests activation of ECs as the key determinant in the pathogenesis of the disease, and NVC may be a useful non-invasive, valid method for assessing the microcirculatory status of children with MIS-C. As a preliminary one, our study may take attention to the use of NVC for follow-up of patients with SARS-CoV-2 infection during clinical course and management.

Contributors

FÇ and NAA designed the form of the study. Examinations of the patients and their follow-up were done by the contribution of all the authors during the study period. They all read and approved the final manuscript. The order of author names is in alphabetical order,

excluding the first author and the corresponding author.

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Data availability statement

The data underlying this article will be shared on reasonable request to the corresponding author.

Declaration of competing interest

The authors declare no conflicts of interest.

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